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Facile Synthesis of Deuterium-Labelled Geranylgeraniols

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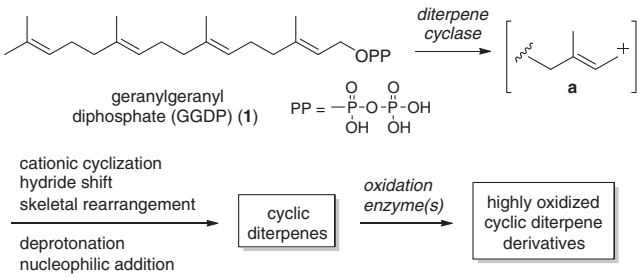
Facile and stereoselective syntheses of three different kinds of deuterium-labelled geranylgeraniol analogs have been achieved. $LiAlD_4$ is used as the deuterium source to ensure high deuterium incorporation. [8,8-d_2]- and [9,9-d_2]-geranylgeraniols have been prepared for the first time. [10-d]-geranylgeraniol was efficiently prepared with a high degree of deuterium incorporation.

Naturally occurring cyclic diterpenes are a structurally diverse class of terpenoids derived from the ubiquitous precursor geranylgeranyl diphosphate (GGDP, **1**) and have received significant attention due to their potential utilities in medicinal and agricultural research areas.[1,2] Cyclic diterpenes are biosynthesized by diterpene cyclases (Scheme 1). The biosynthesis consists of multisteps: an initial activation of GGDP by diterpene cyclases to form highly reactive carbocation **a**, sequential cyclization, cation migrations involving hydride shifts and alkyl group rearrangements, and termination reaction by deprotonation or nucleophilic addition. These reactions are precisely controlled at the reactive site of diterpene cyclases to provide a single cyclic diterpene product. Further functionalization of the product by enzymes, such as oxidase, provides highly oxidized cyclic diterpene derivatives.

Biosynthetic reaction mechanisms of cyclic diterpenes have attracted considerable attention in recent years.[1–16] Many efforts have been devoted to structural and mechanistic studies of diterpene cyclases.[1–16] In these studies, deuterium (D)-labelled geranylgeraniol (GGOH) analogs in which a proton of GGOH (**2**) is replaced with a D have become valuable probes to trace the cation migration with hydride shifts.[17–25] In addition, the D-labelled analogs have been supplied as useful analytical standards for mass spectrometry analysis of the metabolic pathways.[26]



Scheme 1. Biosynthetic pathway of cyclic diterpenes.

The D-labelling of acyclic terpenes, such as GGOH (**2**) and farnesol are divided into three categories: Type 1,[20,21,24] Type 2,[17–19,24,25] and Type 3[22–24] (Figure 1). Previously, syntheses of D-labelled GGOHs have been performed by using 3-oxo-[4,4,4-d_3]-butanoate (**3**), 3-oxo-[2,2-d_2]-butanoate (**4**), $LiAlD_4$, or AlD_3 as the D source. Despite these previous contributions, improved methods with high degrees of D-incorporation, *E*-stereoselectivity, and practical convenience are still required. In this paper, we report efficient and robust syntheses of three different types of D-labelled GGOH analogs, [8,8-d_2]-GGOH (**5**), [10-d]-GGOH (**6**), and [9,9-d_2]-GGOH (**7**), from readily available starting materials. The use of $LiAlD_4$ as the D source ensured efficient D-incorporation into the target analogs **5**–**7**.

The synthesis of [8,8-d_2]-**5** commenced with reduction of compound **8**[27] using $LiAlD_4$ (Scheme 2). A *tert*-butyldiphenylsilyl (TBDPS) group which could be removed with tetrabutylammonium fluoride (TBAF) was chosen as a protecting group of **8** in consideration of the lability of the isoprene unit under acidic conditions. Ester **8** was reduced at −40 to −20 °C to give **9** in 92% yield. When the reaction was performed at elevated temperatures, the undesired 1,4-reduction product was observed as a minor and inseparable by-product. Resultant allyl alcohol **9** was transformed to bromide **10** using the Appel reaction.[28] The bromide **10** was reacted with allylsulfone **11** in the presence of *t*-BuOK to give coupling product **12**. The TBDPS group was then removed with TBAF to give **13** in 67%

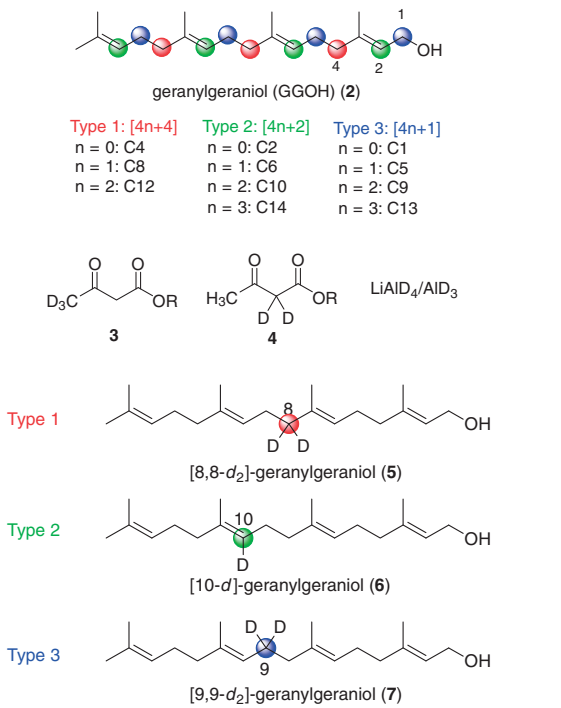
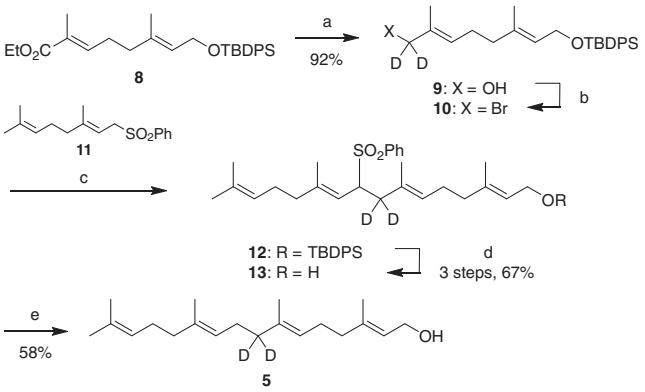
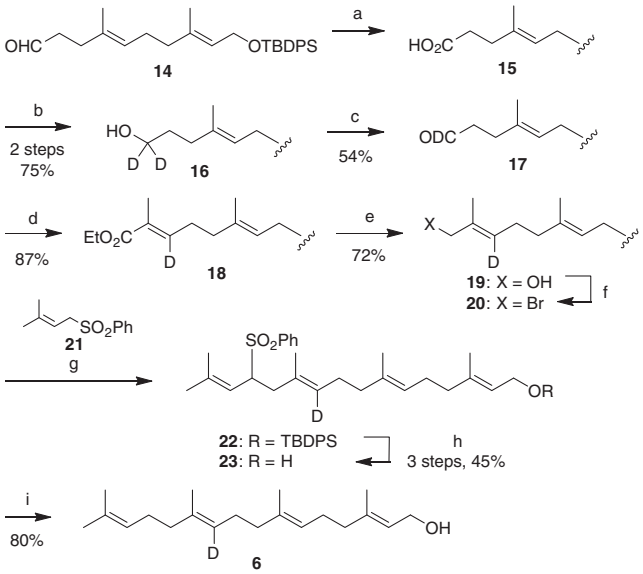


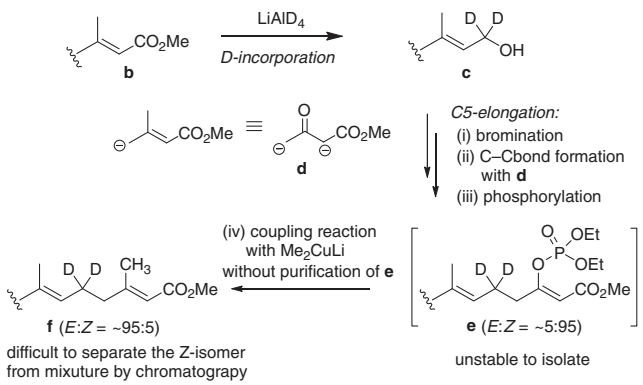
Figure 1. Classification of D-labelling patterns and structures of synthetic targets **5**–**7**.



Scheme 2. Synthesis of Type 1 analog **5**. Reaction conditions: a) $LiAlD_4$ (2.0 equiv), Et_2O, −40 to −20 °C; b) CBr_4 (1.3 equiv), PPh_3 (1.3 equiv), CH_2Cl_2, rt; c) **11** (1.2 equiv), *t*-BuOK (4.0 equiv), THF, −20 °C; d) TBAF (4.0 equiv), THF, rt; e) $LiBHEt_3$ (5.0 equiv), [Pd(dppp)Cl_2] (5 mol %), THF, 0 °C.



Scheme 3. Synthesis of Type 2 analog **6**. Reaction conditions: a) $NaClO_2$ (9.0 equiv), NaH_2PO_4 (7.0 equiv), 2-methyl-2-butene (24 equiv), *t*-BuOH/H_2O, rt; b) $LiAlD_4$ (2.0 equiv), Et_2O, 0 °C; c) PCC (2.0 equiv), CH_2Cl_2, rt; d) ethyl 2-(triphenylphosphoranylidene) propionate (1.5 equiv), CH_2Cl_2, rt; e) $LiAlH_4$ (2.0 equiv), Et_2O, −40 to −20 °C; f) CBr_4 (1.3 equiv), PPh_3 (1.3 equiv), CH_2Cl_2, rt; g) **21** (1.2 equiv), *t*-BuOK (4.0 equiv), THF, −20 °C; h) TBAF (4.0 equiv), THF, rt; i) $LiBHEt_3$ (5.0 equiv), [Pd(dppp)Cl_2] (5 mol %), THF, 0 °C.



Scheme 4. Previous synthesis of Type 3 analog via phosphonate.

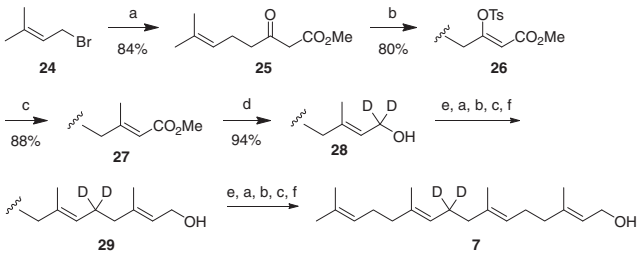
yield over three steps. The phenylsulfonyl group was then removed using $LiBHEt_3$ in the presence of [Pd(dppp)Cl_2] (dppp: 1,3-bis(diphenylphosphino)propane)[29] to give **5** in 58% yield.

In accordance with previous studies to prepare Type 2 analogs,[19,24] we initially attempted the synthesis of **6** using **4** to D-label at position C10. However, in our experiments, **4** was found to be labile and allowed the rapid deuterium–proton exchange at the methylene. To establish the more robust D-incorporation method, we turned our attention to a synthetic route using $LiAlD_4$ (Scheme 3). Known aldehyde **14**[27,30] was oxidized to carboxylic acid **15**, which was then reduced with $LiAlD_4$ to give **16** in 75% over two steps. Alcohol **16** was oxidized to aldehyde **17** using PCC. The Wittig reaction using a stable ylide (ethyl 2-(triphenylphosphoranylidene) propionate) secured the *E*-stereoselective formation of **18** with high yield. Reduction of **18** at low temperature gave **19** in 72% yield. Alcohol **19** was then transformed into **23** in three steps: (i) Appel type bromination,[28] (ii) coupling with sulfone **21**, and (iii) removal of the TBDPS group with TBAF. Desulfonylation of **23** using $LiBHEt_3$/[Pd(dppp)Cl_2] afforded **6** in 80% yield.

Previous syntheses of Type 3 analogs have been performed by the reduction of α,β-unsaturated ester **b** with $LiAlD_4$ to give **c**, followed by C5-chain elongation of **c** using as an isoprene equivalent **d** (Scheme 4).[20–22,31] To remedy the practical drawbacks in regard to the instability of vinyl phosphate **e** and preparation of all *E*-isomers as a single product, we applied an alternative approach via a vinyl tosylate[32] to the stereoselective synthesis of Type 3 analog **7** (Scheme 5).

A dienolate of methyl acetoacetate was reacted with bromide **24** to give **25** in 84% yield (Scheme 5). Ketoester **25** was transformed to *Z*-tosylate **26** under Tanabe's mild conditions in a highly stereoselective manner (95:5).[33,34] *Z*-Tosylate **26** was found to be more stable than vinyl phosphonate **e** and the undesired *E*-vinyl tosylate enabled removal by silica gel column chromatography. The Negishi coupling[35] was performed using commercially available Me_2Zn in the presence of a Pd catalyst at ambient temperature to give **27** as a single stereoisomer. Careful reduction of **27** with $LiAlD_4$ at −40 to −20 °C provided D-labelled alcohol **27** in high yield. Allyl alcohol **28** was then subjected to a series of reactions: (i) bromination using PBr_3, (ii) C–C bond formation with the dianion enolate of methyl acetoacetate, (iii) conversion to *Z*-tosylate, (iv) Negishi cross-coupling, and (v) reduction of the resultant α,β-unsaturated ester at low temperature to give **29**. The same reaction sequence was repeated to give **7** in good overall yield.

In summary, we have developed practical synthetic routes to access different types of D-labelled GGOHs **5**–**7**. ^{1}H NMR and ^{13}C NMR data of **5**–**7** revealed a high D-incorporation ratio (Supporting Information). These methods could be applied to the synthesis of various D-labelled GGOH analogs. Attempts



Scheme 5. Synthesis of Type 3 analog **7**. Reaction conditions: a) Methyl acetoacetate (2.9 equiv), NaH (3.0 equiv), *n*-BuLi (3.0 equiv), THF, −40 to 0 °C; b) TsCl (1.5 equiv), LiCl (5.0 equiv), Et_3N (1.5 equiv), NMI (1.5 equiv), CH_2Cl_2, rt; c) Me_2Zn (2.0 equiv), $[Pd(PPh_3)_2Cl_2]$ (3 mol %), THF, rt; d) $LiAlD_4$ (1.5 equiv), Et_2O, −40 to −20 °C; e) PBr_3 (0.5 equiv), Et_2O, −15 to 0 °C; (f) $LiAlH_4$ (1.5 equiv), Et_2O, −40 to −20 °C.

to elucidate the mechanism of the hydride shift initiated by diterpene cyclases are ongoing.

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Supporting Information

Experimental procedures, compound characterization, copies of ^1H and ^{13}C NMR spectra of **5**, **6**, and **7**. This material is available free of charge on J-STAGE.

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